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The Effect of Gelling Agent Concentration on the Characteristic of Gel Produced From the Mixture of *Semi-refined Carrageenan* and Glukomannan

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Abstract

Carrageenan and konjac glucomannan mixture is synergistic in producing a gel with high gel strength, good and elastic texture, as well as low syneresis. The synergistic effect is a very crucial factor in the utilization of carrageenan and konjac glucomannan mixture extensively for both food and non-food purposes. The use of these two types of polysaccharides can also reduce dependence on imported gelling agents such as locust bean gum. This study aims to produce such concentration of *semi-refined carrageenan* and glucomannan mixture as best gelling agents by evaluating the physical properties of the resulted gel mixture in terms of gel strength, hardness, rigidity, syneresis, and gel micro structure. The experimental design used in this study is completely randomized design with two factors to compare namely proportion ratio of gel-forming material and concentration of gel-forming material. Data is then processed using analysis of variance and the *Tukey's test*.

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The results show that the concentration of gel-forming material 7% in comparison with *semi-refined carrageenan* and glucomannan mixture of 1: 1 has the highest gel strength value of 3361.14 g/cm²; lowest syneresis value of 0.7%, with such dense and compact gel matrix form.

Keywords: Concentration; Semi-refined carrageenan; Glucomannan; Gel Characteristic

1. Introduction

Carrageenan is sulfate galactan specially extracted from red seaweed, such as *Eucheuma* and *Gigartina* [8] and is usually classified into three main groups, namely *kappa-carrageenan*, *iota-carrageenan* and *lambda carrageenan*, each with different chemical structures resulting in gels which also show typical characteristics in KCl salt solution [23,17]. Carrageenan has the ability to form gels in a thermo-reversible way making it widely used as a gelling agent, thickener, and stabilizer in various industries such as food, pharmaceuticals, cosmetics, printing, and textile [29,11]. Carrageenan is typically used as a stabilizer, thickener, gelling, emulsifying, binders and crystallization preventing in the food and beverage, pharmaceutical, and cosmetics industries [23,22,20].

Semi-refined carrageenan (SRC) is one of the carrageenan products with a purity level lower than *refined carrageenan*, as it still contains sulfate groups resulting in a lower gelling ability and is usually found in the forms of powder and chips [16,4].

Konjac glucomannan (KGM) is extracted from the root of the *Amorphophallus konjac* plant [18] and is used as a raw material for food and traditional medicine in China and Japan [12]. Konjac dissolves in both hot water and cold water, with high viscosity and a pH between 4.0 to 7.0; it serves as a gelling agent, thickener, emulsifier, and stabilizer. Based on these properties, it is expected that konjac can be mixed with carrageenan to increase the elasticity of carrageenan [6]. In water, konjac glucomannan takes a highly viscous form. This viscous system can be formed into a gel under a stable heat conditions through heat settings and dilute alkali. Konjac gel is also stable in the presence of acid and salt. Konjac has a synergistic effect with a number of stabilizers, including carrageenan, *xanthan gum*, *locust bean gum*, and starch. The synergistic effect allows the use for various and more functional purposes, and texture for formulation [28,10].

Glucomannan has a property to lower the surface tension of the mixture of kappa carrageenan gel and glucomannan to form a more elastic gel and lower the brittleness property of carrageenan gel so that the gel is made stronger. The mixture of carrageenan and konjac can produce good gels as there is a synergistic relationship in the formation of gels—so as to produce gels with higher gel strength, better texture, and better elasticity [8,10,17,27,21,30,32]. Johnson (2007), as referring to Widjanarko (2008), states that konjac, as a gelling agent, has a unique ability to form reversible and irreversible gels at different conditions. Konjac can form a gel by heating it to 85°C under alkaline conditions (pH 9-10). This gel is heat resistant (irreversible) and remained stable under reheating at a temperature of 100°C and even at a temperature of 200°C [31].

Application and use of semi-refined carrageenan combined with glucomannan in the manufacture of gels and other gel products is still very limited causing data and information about the physical properties of the gel resulted from the combination of the two gelling agents to be very limited. Therefore, it is necessary to conduct a study on the effect of the concentration of gelling agents derived from the combination of the two gelling agents as to provide more comprehensive data related to the physical properties of the gel produced. Specifically, the produced gel is expected to have higher gel strength and lower syneresis for wider formulation purposes both for food and non-food purposes. The purpose of this study is to produce a mixture of *semi-refined carrageenan* and glucomannan as a best gelling agent as well as to evaluate its characteristics.

2. Methods

This study took place from April 2013 to August 2014 at the Laboratory of Basic Applied Sciences Department of Agroindustrial Technology, Faculty of Agricultural Technology, Bogor Agricultural University, Interuniversity Center Laboratory of Food Chemistry, Bogor Agricultural University, and the Zoology Laboratory of Sciences Institute of Indonesia, Cibinong. Materials used in this study consisted of *semi-refined carrageenan* obtained from PT Ocean Fresh, Bandung, West Java; glucomannan obtained from PT Cottonii Sejahtera, Makassar, South Sulawesi; propylene glycol obtained from PT Brataco Cikarang, Bekasi, manufactured by Dow Chemical Pacific, Singapore; and sodium benzoate obtained from PT Setia Guna Bogor; and distilled water. The equipment used in the study consisted of Texture analyzer of the STEVENS-LFRA, SEM Brand (Jeol, JSM-5310LV Japan), electric stove, mercury thermometer, stirrer, spatula, beaker, plastic mold, and analytical balance. The method used in this study was a laboratory experiment method which consisted of the production of gel with the concentration of each gelling agent for each variation of the mixture 1: 1 and 1: 3 (w/w) composed of 5%, 6%, 7% (w/v). The next stage was doing the analysis on several physical parameters consisting of gel strength, rigidity, hardness, syneresis and gel microstructure.

2.1. Gel Strength, Hardness, and Rigidity [13]

The analysis was performed using a Texture Analyzer. The awl used measured 0.1923 cm^2 at a speed of 0.5 mm / sec to a depth of 20 mm; when the awl was in the middle of the gel surface, texture analyzer was activated until the awl penetrated the surface of the gel. Evaluation of the results of measurement was performed by reading the graphics generated. Gel strength is observed as the peak force (g) at the time of gel rupture divided by width of the contact area / pressure field (cm^2). Hardness is the highest peak value (cm) multiplied by the calibration value (g/cm) and gel rigidity value refers to gel strength value (g/cm^2) divided half width of the (cm).

2.2. Syneresis (AOAC) [5]

Syneresis occurs during storage is observed by storing the gel at room temperature for 24 hours. Syneresis is calculated by measuring the weight loss during storage (W_a) and then compared to the initial weight (W_0) of the gel. Syneresis is then expressed by the following formula:

$$\text{Syneresis (\%)} = \frac{W_0 - W_a}{W_0} \times 100\%$$

2.3. Gel matrix [33]

Analysis of the gel matrix was done using *Scanning Electron Microscopy* (SEM). Samples (gel) were cut of 2-3 mm and then prepared (getting the sample in their dry form) and, after going through the process of sample preparation, the gel was then placed on stubs (sample holders) and then coated with gold using a gold sputter coater for 30 minutes with a coating thickness of 400-500 $^{\circ}$ A. Coated samples were placed into the SEM microscope (JEOL, JSM-5310LV Japan) and were then observed at 20 kV of acceleration voltage. The images obtained were recorded and printed.

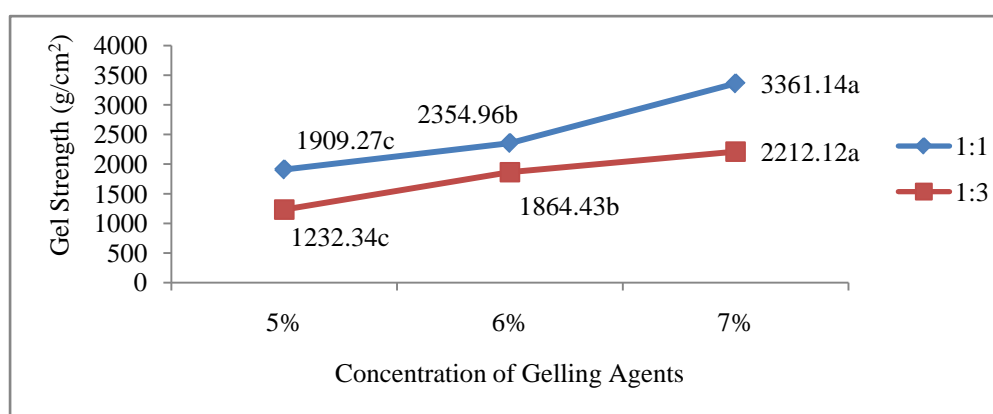
2.4. Design of Experiment and Data Analysis

The experimental design used in this study was completely randomized design with two factors, namely gelling agent ratio and concentration of gelling agent under 3 repeats. Data was analyzed using analysis of variance if the results of the analysis indicate a significant effect then *Tukey's* test would follow to determine which treatment giving significantly different effects on the parameters measured or analyzed [26].

3. Results and Discussion

3.1. Gel Strength

The results of the analysis on gel strength due to examined combined treatment ranged between 1232.34 to 3361.1 g / cm². The results of analysis of variance showed that the ratio of the gelling agents, the concentration of the gelling agents, and the interaction of the gelling agents significantly affected gel strength. *Tukey's* test results showed that the treatment on the ratio of the gelling agents, treatment on the concentration of the gelling agents, and the interaction of the gelling agents and the concentration of the gelling agents were significantly different. The results of the analysis of the gel strength can be seen in Figure 1.



Figures with different superscripts (a,b) represent significant difference ($p < 0.05$)

Figure 1: The effect of gelling agent's concentration on gel strength

Differences in gel strength may be due to further reduction of free water contained in the gel as syneresis occurs. The presence of glucomannan whose function is similar to that of fiber making the free water in the gel absorbed by the molecular structure of glucomannan—in other words, glucomannan has a good ability to absorb water. Increased concentration of glucomannan results in a decrease in gel strength since the texture of the resulted gel is rather mushy so the ability to withstand weight becomes smaller; yet, increased concentration of *semi-refined carrageenan* leads to increasing gel strength because the resulted gel is more dense and compact so it has a good ability to withstand weight per width area.

Temperature above the melting point, kappa carrageenan polymers in solution form a random formation. When it is being cooled, the random formation turns into a double helical chain that allows the formation of cross ties to continuously create a network (matrix). Subsequent cooling causes the polymers to become strongly cross-linked and to form aggregates of a strong gel [14].

Increased gel strength due to the synergistic effect between the glucomannan and carrageenan leads to increased gel strength and decreased syneresis; while decreased gel strength, due to stiffness of the chain such as the number, types, and positions of sulfate, has a very important influence to the formation of the gel, as it will inhibit the formation and accumulation of double helix which further lowers the gel strength [16]. Increased gel strength value is because glucomannan is adsorbed on the surface of the junction zone of the aggregated carrageenan and leads to the amalgamation of carrageenan and glucomannan [2]. After losing water as syneresis, compactness or gel strength increases in line with the effectiveness of hydrocolloid concentration [7].

The addition of glucomannan proportion and concentration will affect the texture of the material such as hardness and elasticity [34,16].

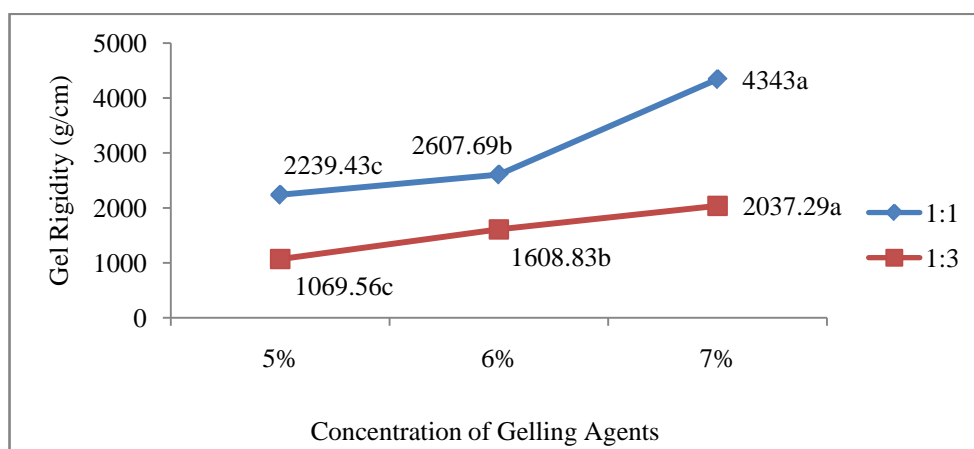
3.2. Gel Rigidity

The results of the analysis on gel rigidity due to examined combined treatment ranged between 1069.56 to 4343 g / cm. The results of analysis of variance showed that the ratio of the gelling agents, the concentration of the gelling agents, and the interaction of the gelling agents significantly affected gel rigidity. *Tukey's* test results showed that the treatment on the ratio of the gelling agents, treatment on the concentration of the gelling agents, and the interaction of the ratio of the gelling agents and the concentration of the gelling agents were significantly different. The results of the analysis of the gel rigidity can be seen in Figure 2.

Increased ratio and concentration of *semi-refined carrageenan* will affect gel rigidity because the resulted gel has a dense, compact, and hard texture due to evaporation of free water in the gel, causing shrinkage in the gel; in contrast, increased ratio and concentration of glucomannan will produce a softer gel because glucomannan has a good ability to absorb water.

Gel rigidity and structuring is influenced by diffusion process of small molecules, further stated that increased concentration of carrageenan also reduces the amount of free water and reduces the mobility of small-sized

molecules. Kappa carrageenan has a rigid gel type or fragile, characterized by high syneresis, that is flow of fluid on the surface of the gel [24].



Figures with different superscripts (a,b) represent significant difference ($p < 0.05$)

Figure 2: The effect of gelling agent's concentration on gel rigidity

The addition of glucomannan ratio and concentration will affect the texture of the material such as hardness and elasticity [34,16]. During the cooling process, carrageenan polymers form the double helix structure that results in junction points of the polymer chains and cavities in between the chains. Glucomannan chains will fill cavities between kappa carrageenan chains. The more the content of glucomannan, the more carrageenan cavities will be filled with viscous and pseudoplastic solution, and so the gel becomes even more dense and elastic [16]. Further noted, variations in the composition of the gel indicate that fluid released is in accordance with the coefficient of thermal retraction, which depends on the elasticity [3].

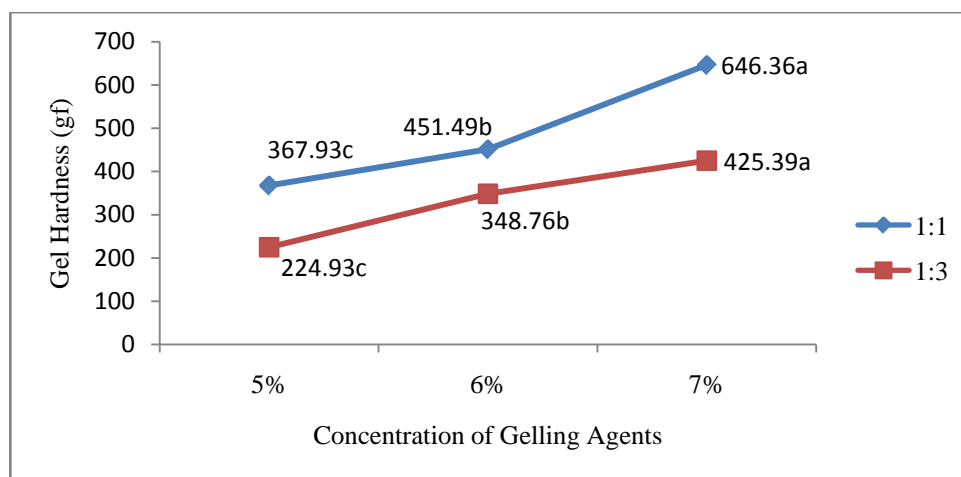
3.3. Gel Hardness

The results of the analysis on gel hardness due to examined combined treatment ranged between 224.93 to 646.36 gf. The results of analysis of variance showed that the ratio of the gelling agents, the concentration of the gelling agents, and the interaction of the gelling agents significantly affected gel hardness. *Tukey's* test results showed that the treatment on the ratio of the gelling agents, treatment on the concentration of the gelling agents, and the interaction of the ratio of the gelling agents and the concentration of the gelling agents were significantly different. The results of the analysis of the gel hardness can be seen in Figure 3.

Increased ratio and concentration of *semi-refined carrageenan* will affect gel hardness because the resulted gel has a dense and hard texture due to the formation of double strands followed by cross-link producing nets with strong ties. In contrast, increased addition of glucomannan ratio and concentration will produce a more mushy

gel because glucomannan has a good ability to absorb water, so the amount of free water presenting in the gel increased.

The addition of glucomannan ratio and concentration will affect the texture of the material such as hardness and elasticity [34,16]. The high number of junction zones may be one reason for the high rate of syneresis. More junction zones can lead to increased syneresis, due to the formation of helix and aggregates that continue to occur during storage so gel bonds shrink and free up more water causing gel texture to become harder [19].



Figures with different superscripts (a,b) represent significant difference ($p < 0.05$)

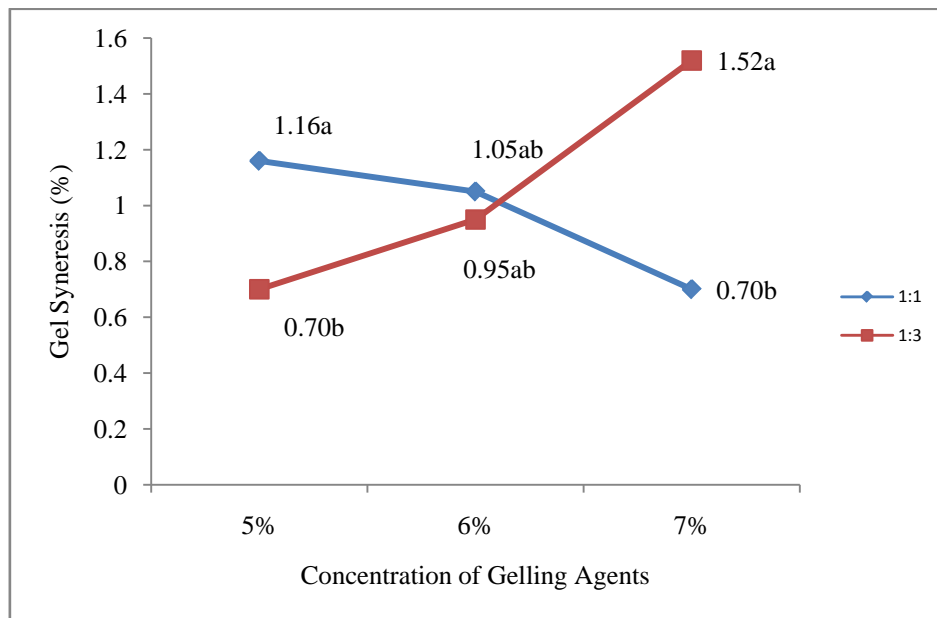
Figure 3: The effect of gelling agent's concentration on gel hardness

3.4. Gel Syneresis

The results of the analysis on gel syneresis due to examined combined treatment ranged between 0.7 to 1.52%. The results of analysis of variance showed that the ratio of the gelling agents and the concentration of the gelling agents did not significantly affect gel syneresis, yet the interaction of the gelling agents significantly affected gel syneresis. *Tukey's* test results showed that the treatment on the ratio of the gelling agents and the treatment on the concentration of the gelling agents were significantly different with gel syneresis, yet the interaction of the ratio of the gelling agents and the concentration of the gelling agents were significantly different with gel syneresis. The results of the analysis of the gel syneresis can be seen in Figure 4.

The syneresis value of *semi-refined carrageenan* combined with *locust bean gum* at a concentration of 1.5% is 0.90% [25]; Syneresis values ranged from 8.64 to 17.56% [30]. The combination of carrageenan and glucomannan at a concentration of 5% in the ratio of the gelling agent of 60:40 produce high gel strength and low syneresis [15].

Aggregate formation continues during storage, and this may be the cause of syneresis. Aggregate formation causes the gel to shrink so it tends to squeeze the water out of the cell [16]. Of the three types of carrageenan—kappa, iota, and lambda only kappa carrageenan undergoes syneresis if it is in the form of gel [17].



Figures with different superscripts (a,b) represent significant difference ($p < 0.05$)

Figure 4: The effect of gelling agents' concentration on gel syneresis

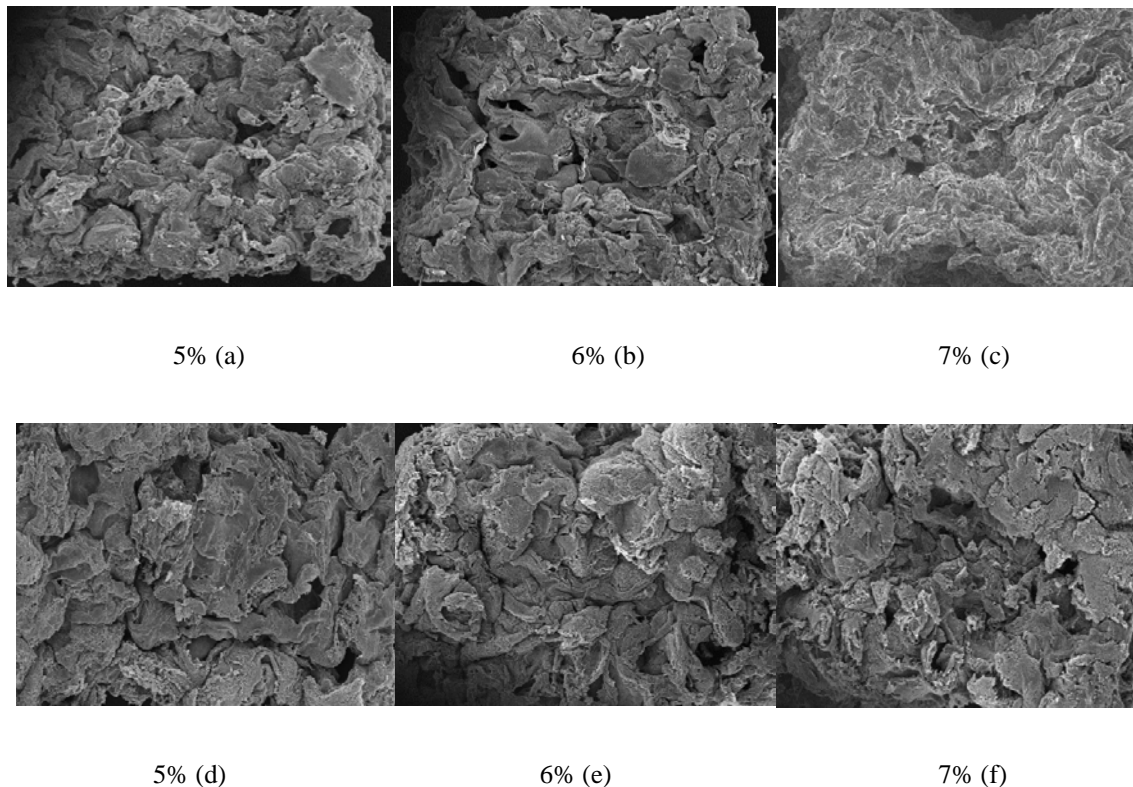
Syneresis of water in the gel is a natural phenomenon, in which excessively free water is sent out of the gel matrix. These events can be minimized by determining the appropriate proportion and concentration of hydrocolloid materials and the addition of supporting gelling agents [7]. Gel syneresis is affected by temperature, humidity, concentration of salt (KCl) and concentration of polysaccharides [3].

3.5. Gel Micro Structure

The results of the observation on the gel matrix structure of each concentration of gelling agent with the proportion of gelling agents of 1: 1 and 1: 3 by using SEM (*Scanning Electron Microscopy*) can be seen in Figure 5.

Based on Figure 5 (a, b, c), it can be seen that the gel matrix structure for a concentration of 5% (a) has a lot of folds / clots and is overlapped; for a concentration of 6% (b), it has a slight folds / clots with some parts looking somewhat dense and compact; while for a concentration of 7% (c), it has a more compact and dense gel matrix.

During the cooling process, carrageenan polymers form the double helix structure that results in junction points of the polymer chains and cavities in between the chains. Glucomannan chains will fill cavities between kappa carrageenan chains making the gel increasingly dense and compact [16].



Note on figures: Type JSM-5000, MAG X35, ACCV 20 kV, WIDTH 3.77 mm

Figure5: Gel micro structure of a combination of *semi-refined carrageenan* and glucomannan on various concentrations with two ratios of 1:1 and 1:3

Furthermore, based on Figure 5 (d, e, f), it can be seen that the structure of the gel matrix to a concentration of 5% and 6% is almost similar, with clots and folds, and is separated; while at a concentration of 7%, it has a more compact structure with little clots.

The function of glucomannan that is similar to fiber causes water to be absorbed into glucomannan molecules, thereby increasing the ability of glucomannan to bind water [14,27]. The addition of glucomannan will affect the texture of the material such as hardness and elasticity [34].

4. Conclusion

Concentration of the gelling agents at 7% in a ratio of 1: 1 of the gelling agents produces the following physical properties of the gel mixture of *semi-refined carrageenan* and glucomannan: gel strength of 3361.14 g / cm²; gel

rigidity of 4343 g / cm; gel hardness of 646.36 gf; gel syneresis of 0.7%; dense and compact gel matrix structure.

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